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Ames Research Center



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Stabilization of Lactate Dehydrogenase

The problem:

Many enzymes are unstable and exhibit a tendency to denature upon storage, even at low temperatures. Thus, there is a need to develop methods for preserving enzymes in a stable and fully active form.

The solution:

Addition of substances such as dimethylsulfoxide, glycerol, and gelatin to aqueous solutions of the lactate dehydrogenase; the enzyme maintains full activity when stored 19 months at 2 to 6°C, whereas it has only 50% of its activity at the end of one month in the absence of stabilizers.

How it's done:

The criterion of stability of beef heart lactate dehydrogenase solutions is based on observation that thermal inactivation at 37°C appears to be a function of incubation time at selected conditions of enzyme concentration, pH, and buffer ionic strength. Normally, approximately 30 minutes is required for 50% reduction of lactate dehydrogenase activity. The extent to which the enzyme is protected from 50% thermal inactivation is thus a measure of the efficacy of the additive under study. The stabilizing and labilizing effects of a series of diverse additives have been evaluated by this test.

It has been found that dilute aqueous solutions of the dehydrogenase are stabilized by additions of various organic additives. Dimethylsulfoxide is especially effective in concentrations of 20 to 40 percent by weight. For example, in the presence of 40-percent dimethylsulfoxide, beef heart lactate dehydrogenase retained about 65 percent of its original

activity after storage for 20 months at $4 \pm 2^\circ\text{C}$. Lower concentrations are less effective; in the presence of 20-percent dimethylsulfoxide, all activity is lost in 16 months.

The stabilizing property of dimethylsulfoxide suggested a study of other additives, and it was found that the inclusion of gelatin enhanced stability still further. For example, beef heart lactate dehydrogenase retains virtually all of its activity when stored 19 months in aqueous 40-percent dimethylsulfoxide solutions containing 0.5 percent gelatin; under similar storage conditions, aqueous solutions retain only 10 percent of the initial activity.

Glycerol, ethyl, and methyl alcohol, isobutanol, etc. are also useful stabilizers; bovine serum albumin may be used in place of gelatin. However, each stabilizing system has to be evaluated for the particular enzyme that is to be preserved.

Reference:

George, H.; McMahan, J.; Bowler, K.; and Elliott, M.: Stabilization of lactate and malate dehydrogenase by organic solvents. *Biochimica et Biophysica Acta*, vol. 191, page 466, 1969.

Notes:

1. The following documentation may be obtained from:

National Technical Information Service
Springfield, Virginia 22151
Single document price \$3.00
(or microfiche \$0.95)
Reference: NASA CR-73460 (N70-31057),
Studies on Stabilization of Enzymes.

(continued overleaf)

2. No additional documentation is available. Specific questions, however, may be directed to:

Technology Utilization Officer
Ames Research Center
Moffett Field, California 94035
Reference: B72-10062

Patent status:

No patent action is contemplated by NASA.

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